Digestible and metabolizable energy content of crude glycerin originating from different sources in nursery pigs[1]

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ABSTRACT: The energy value of crude glycerin from different biodiesel production facilities was determined in nursery pigs (initial BW of 10.4 kg) to predict apparent DE and ME based on the composition of crude glycerin. Dietary treatments consisted of a basal diet, or diets containing crude glycerin from various biodiesel production facilities supplemented in the diet at approximately 9.1%. Because of bulk density differences, 2 glycerin products were supplemented at either 7.7 or 6.9%. In addition, soybean oil and lard were included at 6.7% as 2 dietary treatments to serve as positive controls. Each diet was fed twice daily to pigs in individual metabolism crates. After a 6-d adjustment period, a 4-d balance experiment was conducted. During the collection period, feces and urine were collected daily and stored at 0°C until analysis. The GE of each test ingredient and diet and of urine and fecal samples from each pig were determined by isoperibol bomb calorimetry. The DE and ME values of crude glycerol were estimated by difference, whereby the DE and ME content of the basal diet was subtracted from the complete diet containing the test ingredient. Gross energy, DE, and ME of US Pharmacopeia grade glycerin were determined to be 4,325, 4,457, and 3,682 kcal/kg, respectively. In contrast, GE of the crude glycerin samples ranged from 3,173 to 6,021 kcal/kg, DE ranged from 3,022 to 5,228 kcal/kg, and ME ranged from 2,535 to 5,206 kcal/kg, reflecting the content of glycerol, methanol, and FFA in the crude glycerin. The GE, DE, and ME of soybean oil and lard were determined to be 9,443, 8,567, and 8,469 kcal/kg, and 9,456, 8,524, and 8,639 kcal/kg, respectively. The stepwise regression prediction of the ME in crude glycerin exhibited R^2 of only 0.41 [ME, kcal/kg (as-is basis) = (37.09 × % of glycerin) + (97.15 × % of fatty acids)], whereas prediction of GE achieved an R^2 of 0.99 [GE, kcal/kg (as-is basis) = −236 + (46.08 × % of glycerin) + (61.78 × % of methanol) + (103.62 × % of fatty acids)]. On average, the ME of crude glycerin was 85.4% of its GE (SE 5.3) and did not differ by glycerin source. The data provided in these experiments indicate that crude glycerin is a valuable energy source, with its GE concentration dependent on the concentration of glycerin, methanol, and fatty acids, and with ME as a percentage of GE averaging 85.4%.

Key words: biofuel, crude glycerin, digestible energy, metabolizable energy, pig

INTRODUCTION

Biodiesel can be produced by a variety of esterification technologies using vegetable oil, animal fat, or yellow grease as the initial feedstock (Ma and Hanna, 1999; Van Gerpen, 2005; Thompson and He, 2006). Production of biodiesel from oils other than soybean oil has increased because of the recent increase in the price of soybean oil. With approximately 80 g of crude glycerin generated for every liter of biodiesel produced, the potential crude glycerin generated from biodiesel production is large (National Biodiesel Board, 2007). Glycerin is readily absorbed by the gastrointestinal tract (Tao et al., 1983), and the metabolism of glycerol has been reviewed previously (Lin, 1977; Brisson et al., 2001).

Research evaluating the ME value of glycerin in nonruminants is limited (Bartelt and Schneider, 2002; Dozier et al., 2008, Lammers et al., 2008a,c). In swine, Bartelt and Schneider (2002) used pure glycerin in diets

Table 1. Chemical analysis (%, as-is basis) of crude glycerin samples[1]

Sample[2]	Source[3]	Glycerin	Moisture	Methanol	pH	NaCl	Ash	Fatty acids
USP	—	99.62	0.348	ND	5.99	0.013	0.01	0.02
ADM-MO	SB	83.88	10.161	0.0059	6.30	5.997	5.83	0.12
AGP-IA	SB	83.49	13.397	0.1137	5.53	2.838	2.93	0.07
REG-MN	SB	85.76	8.347	0.0260	6.34	6.065	5.87	ND
REG-R	SB	83.96	9.363	0.0072	5.82	6.346	6.45	0.22
REG-WL	SB	84.59	9.201	0.0309	5.73	6.000	5.90	0.28
WW-TX	SB	81.34	11.406	0.1209	6.59	6.577	7.12	0.01
WW-OH	TA	73.34	24.367	0.0290	3.99	0.073	1.91	0.04
IW-AC	YG	93.81	4.071	0.0406	6.10	0.162	1.93	0.15
IW-NA	YG	52.79	4.157	3.4938	8.56	1.977	4.72	34.84
USB-GA	PF	51.54	4.989	14.9875	9.28	0.011	4.20	24.28

[1]Samples analyzed as described in Lammers et al. (2008b); courtesy of Ag Processing Inc. (Omaha, NE). Glycerin content determined by difference as 100 − % of methanol − % of total fatty acid − % of moisture − % of ash. ND = not detected.

[2]USP = US Pharmacopeia grade glycerin; ADM-MO = Archer Daniels Midland (Mexico, MO); AGP-IA = Ag Processing Inc. (Sergeant Bluff, IA); REG-MN = Renewable Energy Group (Albert Lea, MN); REG-R = Renewable Energy Group (Ralston, IA); REG-WL = Renewable Energy Group (Wall Lake, IA); WW-TX = Westway Feed Products (Houston, TX); WW-OH = Westway Feed Products (Cincinnati, OH); IW-AC = Imperial Western Products acidulated glycerin (Coachella, CA); IW-NA = Imperial Western Products nonacidulated glycerin (Coachella, CA); USB-GA = US Biofuels (Rome, GA).

[3]Feedstock source, which was obtained by the biodiesel plant for biodiesel production. SB = soybean oil from a hexane soybean crush plant, except for REG-R, for which the soybean oil was obtained from extruded soybeans; TA = tallow; YG = yellow grease; PF = poultry fat.

containing up to 15% glycerin and reported an average ME of 3,525 kcal/kg. Recently, Lammers et al. (2008c) reported that a crude glycerin containing 87% glycerin obtained from a biodiesel facility using soybean oil as the initial feedstock contained 3,207 kcal/kg when fed to starting and finishing pigs, a value that is only slightly greater than the value obtained by Bartelt and Schneider (2002) on an equivalent glycerin basis. Because it is not known whether the variation in crude glycerin among biodiesel production facilities would affect its DE or ME value in swine, a balance experiment was designed to determine the DE and ME concentrations of various crude glycerin samples obtained from different production facilities. From these data, an equation estimating the ME concentration was generated based on the composition of the crude glycerin. A second growth experiment was conducted to evaluate the ability of growing pigs to gain BW efficiently (BW gain:Mcal of ME) on these diets, regardless of the glycerin source.

MATERIALS AND METHODS

The Iowa State University Animal Care and Use Committee approved all experimental protocols.

General Pig Management

Crude glycerin was obtained from various biodiesel plants using soybean oil (both hexane and extruded soybean processing), tallow, yellow grease, or poultry fat. The crude glycerol was characterized courtesy of Ag Processing Inc. (Omaha, NE) by the standard techniques used at biodiesel plants, as described previously (Lammers et al., 2008b). The initial biodiesel feedstock source and composition of each crude glycerin used in Exp. 1 and 2 are detailed in Table 1.

Dietary treatments consisted of a common basal diet, which met or exceeded the NRC (1998) requirements (Table 2), or diets containing 9.09% crude glycerin (**USP** = US Pharmacopeia grade glycerin; **ADM-MO** = Archer Daniels Midland, Mexico, MO; **AGP-IA** = Ag Processing Inc., Sergeant Bluff, IA; **REG-MN** = Renewable Energy Group, Albert Lea, MN; **REG-R** = Renewable Energy Group, Ralston, IA; **REG-WL** = Renewable Energy Group, Wall Lake, IA; **WW-OH** = Westway Feed Products, Cincinnati, OH; **WW-TX** = Westway Feed Products, Houston, TX; **IW-AC** = Imperial Western Products acidulated glycerin, Coachella, CA), 7.72% crude glycerin (**IW-NA** = Imperial Western Products nonacidulated glycerin, Coachella, CA), or 6.91% crude glycerin (**USB-GA** = US Biofuels, Rome, GA). The differences in addition of some products were due to their bulk density and our ability to liquefy the product before mixer addition because both IW-NA and USG-GA were solid at room temperature. Products obtained were from hexane-derived soybean oil (AGP-IA, REG-WL, REG-MN, ADM-MO, and WW-TX), expeller-derived soybean oil (REG-R), tallow (WW-OH), yellow grease (IW-AC and IW-NA), or poultry fat (USB-GA). We also chose to obtain a product from 1 plant before acidulation (IW-NA) that had an elevated FFA content. In addition, the product from a plant using poultry fat (USB-GA) contained a greater content of fatty acids. We also included an evaluation of soybean oil and lard, added at 6.69% of the diet, which were used as high-energy controls, to validate our methodology, as outlined by Adeola (2001). We chose moderate inclusion levels for all products used because there was some concern about the effect of glycerin inclusion level on ME determination (Bartelt and Schneider, 2002; Lammers et al., 2008c) and on the ability of the mixed feed to flow in feeding systems (Cerrate et al., 2006).

Table 2. Composition of basal diet (as-is basis)

Item	Amount, %
Ingredient	
Corn	57.49
Soybean meal, 46.5% CP	24.50
Dried whey	10.00
Fish meal, select	5.00
Plasma, Appetein[1]	1.25
Soybean oil	0.15
Defluorinated phosphate	0.61
Limestone	0.14
Sodium chloride	0.35
Vitamin mix[2]	0.30
Trace mineral mix[3]	0.20
DL-Methionine	0.01
Calculated composition	
ME, kcal/kg	3,325
CP	21.00
True ileal digestible Lys	1.15
Calcium	0.74
Phosphorus$_{available}$	0.40

[1]American Protein Corp. (Ankeny, IA).

[2]Provided the following per kilogram of diet: vitamin A, 6,614 IU; vitamin D_3, 1,653 IU; vitamin E, 33 IU; vitamin B_{12}, 33 µg; riboflavin, 10 mg; niacin, 50 mg; D-pantothenic acid, 26 mg.

[3]Provided the following per kilogram of diet: Zn, 225 mg as ZnO; Fe, 263 mg as Fe_2SO_4; Cu, 26 mg as CuO; Mn, 90 mg as MnO_2; I, 3.0 mg as CaI; Se, 0.3 mg as Na_2SeO_3.

Table 3. Daily allowance of basal and test supplement in pigs fed various energy-containing feedstuffs over the 4-d balance experiment (as-is basis)

Diet[1]	Control diet,[2] g	Test supplement,[3] g (%)
Control	500	—
USP	455	46 (9.09)
ADM-MO	455	46 (9.09)
AGP-IA	455	46 (9.09)
REG-MN	455	46 (9.09)
REG-R	455	46 (9.09)
REG-WL	455	46 (9.09)
WW-TX	455	46 (9.09)
WW-OH	455	46 (9.09)
IW-AC	455	46 (9.09)
IW-NA	462	39 (7.72)
USB-GA	466	35 (6.91)
Soybean oil	467	34 (6.69)
Lard	467	34 (6.69)

[1]Control diet or source of glycerin or lipid: USP = US Pharmacopeia grade glycerin; ADM-MO = Archer Daniels Midland (Mexico, MO); AGP-IA = Ag Processing Inc. (Sergeant Bluff, IA); REG-MN = Renewable Energy Group (Albert Lea, MN); REG-R = Renewable Energy Group (Ralston, IA); REG-WL = Renewable Energy Group (Wall Lake, IA); WW-TX = Westway Feed Products (Houston, TX); WW-OH = Westway Feed Products (Cincinnati, OH); IW-AC = Imperial Western Products acidulated glycerin (Coachella, CA); IW-NA = Imperial Western Products nonacidulated glycerin (Coachella, CA); USB-GA = US Biofuels (Rome, GA).

[2]Refers to amount of complex basal diet offered to the pig.

[3]Refers to the amount of test supplement mixed with the amount of basal diet shown in the proceeding column. Numbers in parentheses represent the percentage of test supplement relative to the total feed offered.

Exp. 1

In Exp. 1, three groups of 56 barrows (average initial BW of 10.4 kg, Cambrough 22 females × L337 sires) were randomly assigned to individual metabolism crates (0.53 × 0.71 m) equipped with screens and trays that allowed for total but separate collection of feces and urine. Pigs were randomly assigned to dietary treatments after pen assignment. Pigs were offered 500 g of feed per day, distributed in 2 equal daily meals (Table 3), with feed consumption and refusals recorded at the end of the experimental period. Water was available from a nipple waterer at all times. A 6-d adjustment period was used to adapt the pigs to the metabolism crate and to the dietary treatment before the 4-d total fecal and urine collection period. During the collection period, urine was collected once daily into plastic buckets containing 25 mL of 6 N HCl and was stored at 0°C until the end of the collection period. At the end of the collection period, urine was thawed and weighed, and a subsample was collected and stored at 0°C until subsequent analysis. Feces were also collected daily and stored at 0°C.

Exp. 2

In Exp. 2, two groups of 56 pigs (initial BW of 8.7 kg; Cambrough 22 females × L337 sires), representing 4 replicates of gilts and 4 replicates of barrows, were randomly assigned to individual pens (0.7 × 1.22 m) and then to the same dietary treatments as in Exp. 1. Because of diet availability, the performance study lasted either 25 d (group 1) or 18 d (group 2). Feed and water were offered ad libitum, and pigs and feeders were weighed at the beginning and end of the experiment to determine ADG, ADFI, and G:F.

Chemical Analyses

Feed samples were ground through a 1-mm screen before energy determination. Fecal samples were thawed, dried at 70°C for 48 h, and weighed to determine DM content. Fecal samples were ground through a 1-mm screen in preparation for energy determination. For urine energy determination, 2 mL of urine was added to 0.5 g of dried cellulose and subsequently dried at 50°C for 24 h before energy determination. The GE of feed, feces, and urine plus cellulose was determined using an isoperibol bomb calorimeter (model 1281, Parr Instrument Co., Moline, IL), with benzoic acid used as a standard. Duplicate analyses were performed on all diets and fecal samples from each pig, whereas triplicate analyses were performed on urine plus cellulose from each pig. Urinary energy was determined by subtracting the energy contained in cellulose from that in the urine plus cellulose.

Calculations and Statistical Analysis

Gross energy intake was calculated by multiplying the GE value of the diet fed by feed intake over the 4-d collection period. Apparent DE values were calcu-

lated by subtracting fecal energy from intake energy, and apparent ME values were calculated by subtracting urinary energy from apparent DE. The apparent DE and ME values of the test ingredient were estimated by difference from the basal diet, as described by Adeola (2001). In Exp. 1, observations from 151 of the 168 pigs assigned to dietary treatments were used for analysis. Observations from 17 pigs were not possible to quantify because of diarrhea, constipation, or feed refusal such that any data obtained from these pigs were considered outliers. All 112 pigs were used for analysis in Exp. 2. The individual pig was used as the experimental unit, and data from each experiment were subjected to ANOVA with group and treatment in the model (SAS Inst. Inc., Cary, NC), with reported means being least squares means. Differences among sources of glycerin, fats, and the control diet were tested using contrast statements. Pooled SE was calculated by averaging the SE calculated by the GLM procedure of SAS for the variable of interest. In addition, a stepwise regression model was used to equate the effect of glycerin composition on apparent ME in Exp. 1, with variables having P-values <0.15 maintained in the model.

RESULTS AND DISCUSSION

Compositional variation in crude glycerin (Table 1) was expected. Glycerin content in the soybean oil-based glycerin products was relatively consistent, averaging 83.9%. For crude glycerin obtained from tallow (WW-OH), glycerin content was less than in other crude glycerin products, whereas its water content was greater. Products with greater FFA content (IW-NA and USB-GA) also had less glycerin content. As expected, acidulation increased glycerin content at the expense of fatty acid content (IW-AC vs. IW-NA). Crude glycerin is a viscous liquid and all samples were viscous except for WW-OH, which contained increased water content, and IW-NA and USB-GA, which were solid at room temperature because of their elevated FFA content. In general, all other samples contained small amounts of FFA, which are indicative of efficient fatty acid esterification at the biodiesel production facility. Only 2 samples, IW-NA and USG-GA, had an elevated content of fatty acids. This was expected for the IW-NA sample because it was not acidulated, whereby the soap stock is acidified, reducing its emulsifying properties such that the amount of fatty acids remaining in the crude glycerin is reduced (Ma and Hanna, 1999; Van Gerpen, 2005; Thompson and He, 2006). We cannot explain why the USB-GA sample contained an elevated content of fatty acids because no additional information was available from the manufacturer. Likewise, methanol content was relatively small in all samples except for the IW-NA and USB-GA samples, which contained 3.49 and 14.99% methanol, respectively. Recovery of methanol by a biodiesel plant also relates to the production efficiency of the plant because the methanol recovered is reused in the biodiesel process (Ma and Hanna, 1999;

Van Gerpen, 2005; Thompson and He, 2006). Because the IW-NA was not a final product from this location, the increased methanol content was not unexpected because the final product, which is acidulated (IW-AC), contained a small content of methanol. Aside from the cost of methanol for the biodiesel production facility, methanol content is a concern in crude glycerin fed to livestock, as described in detail by Lammers et al. (2008b,c). All but USP, WW-OH, IW-AC, and USB-GA contained moderate amounts of ash, which, in most cases, was NaCl. We expect that the 3 products with increased ash but reduced NaCl (WW-OH, IW-AC, and USB-GA) were from biodiesel facilities using a K-based instead of an Na-based catalyst. If elevated content of crude glycerin were to be supplemented in the diet, the level of NaCl or KCl should be considered and balanced accordingly (Lammers et al., 2008b).

In Exp. 1, the removal of 17 pigs for various reasons was unexpected because the herd health at the Iowa State University swine farm was normal during both experiments (Exp. 1, January through March; Exp. 2, March through April). It was noted, however, that several pigs exhibited a fair number of loose stools or feed refusals in Exp. 1, but this could not be attributed to any particular diet. This could be due to a lack of antibiotics in the basal diet (Table 2), but pigs in Exp. 2 did not exhibit this same condition and were fed the same diets. In addition, in past experiments we have not used an antibiotic in the basal diet and have not experienced these problems (Lammers et al., 2008c). During Exp. 1, however, we observed that pigs were noticeably excitable when moved to the metabolism crates and during the entire experiment. This occurred even though each cage allows visual access to other pigs, a pen toy is included in each crate, and pigs were managed appropriately during the experiment. We have no explanation for the excitability of pigs used in Exp. 1, but this may have contributed to the looseness of stools in some of the pigs and, consequently, to the level of variation noted in the experimental results.

Caloric values of the various ingredients are shown in Table 4. For comparative purposes, the ME determined for the basal diet was 3,352 kcal/kg, which is similar to the calculated value of 3,325 kcal/kg based on NRC (1998) requirements. Similarly, the DE and ME determined for soybean oil (8,567 and 8,469 kcal/kg, respectively) and lard (8,524 and 8,639 kcal/kg, respectively) in Exp. 1 are close to those for soybean oil (8,750 and 8,400 kcal/kg, respectively) and lard (8,285 and 7,950 kcal/kg, respectively), as reported in the NRC (1998) requirements, where DE was predicted from FFA concentration and the unsaturated:saturated ratio and ME was predicted as 96% of the DE (Powles et al., 1995). In Exp.1, ME as a percentage of DE for soybean oil and lard (data not shown) averaged 99.85%, which is similar to that reported by Powles et al. (1995). Collectively, these comparisons provided confidence in our balance techniques and the use of the difference method (Adeola, 2001) for determination of DE and ME in these

Table 4. Energy values of various crude glycerin products and lipids in starting swine, Exp. 1[1]

Item	n[2]	GE, kcal/kg	DE, kcal/kg	ME kcal/kg	ME % of GE
Diet[3]					
Control	11	3,945	3,469	3,352	85.0
USP	11	4,325	4,457	3,682	85.2
ADM-MO	10	3,627	3,928	3,389	93.4
AGP-IA	11	3,601	3,022	2,535	70.5
REG-MN	10	3,676	3,789	3,299	89.9
REG-R	11	3,670	3,517	3,024	82.5
REG-WL	11	3,751	3,690	3,274	87.3
WW-TX	11	3,489	3,815	3,259	93.5
WW-OH	10	3,173	3,128	2,794	88.0
IW-AC	11	4,153	3,919	3,440	82.9
IW-NA	10	6,021	5,228	5,206	86.6
USB-GA	12	5,581	4,336	4,446	79.7
Soybean oil	10	9,443	8,567	8,469	89.8
Lard	10	9,456	8,524	8,639	91.2
Pooled SE[4]			239	249	5.3
Contrast,[5] P-value					
Control vs. all others		—	0.01	0.01	0.83
Control vs. USP		—	0.01	0.34	0.98
Control vs. soybean oil and lard		—	0.01	0.01	0.40
USP vs. soybean oil and lard		—	0.01	0.01	0.41
USP vs. soybean oil-based glycerin		—	0.01	0.04	0.86
Soybean oil- vs. tallow-based glycerin		—	0.06	0.22	0.75
Soybean oil- vs. yellow grease-based glycerin		—	0.24	0.24	0.56
Tallow- vs. yellow grease-based glycerin		—	0.02	0.07	0.49

[1]Pigs were adapted to diets and feeding regimens for 6 d before a 4-d collection period. Initial and final BW were 10.4 and 12.8 kg, respectively.

[2]Number of observations (pigs) per dietary treatment.

[3]Control diet or source of glycerin or lipid: USP = US Pharmacopeia grade glycerin; ADM-MO = Archer Daniels Midland (Mexico, MO); AGP-IA = Ag Processing Inc. (Sergeant Bluff, IA); REG-MN = Renewable Energy Group (Albert Lea, MN); REG-R = Renewable Energy Group (Ralston, IA); REG-WL = Renewable Energy Group (Wall Lake, IA); WW-TX = Westway Feed Products (Houston, TX); WW-OH = Westway Feed Products (Cincinnati, OH); IW-AC = Imperial Western Products acidulated glycerin (Coachella, CA); IW-NA = Imperial Western Products nonacidulated glycerin (Coachella, CA); USB-GA = US Biofuels (Rome, GA).

[4]Pooled SE was calculated by averaging the SE error calculated by the GLM procedure (SAS Inst. Inc., Cary, NC) for the variable of interest.

[5]Preplanned contrasts: soybean oil-based glycerin (ADM-MO, AGP-IA, REG-MN, REG-R, REG-WL, WW-TX), tallow-based glycerin (WW-OH), and yellow grease-based glycerin (IW-AC, but excluding IW-NA because of its elevated content of FFA).

products. Specific comparisons of DE or ME values among glycerin products were not relevant for this experiment because the objective was not to compare DE or ME values among products, but to equate ME relative to crude glycerin composition. For example, comparison of the tallow-based glycerin (WW-OH) with either soybean oil-based glycerin (DE, $P = 0.06$; ME, $P = 0.22$) or yellow grease-based glycerin (IC-AC; DE, $P = 0.02$; ME, $P = 0.07$), as shown in Table 4, is not a relevant comparison, given the compositional differences in the respective crude glycerin samples provided in Table 1. We were somewhat surprised with the 2,535 kcal of ME/kg determined for AGP-IA, which appeared to be less than expected, given that we had previously obtained a sample from this same plant and determined an ME value of 3,207 kcal/kg (Lammers et al., 2008c). However, it should be noted that the composition of samples did vary [glycerin (83.49 vs. 86.95%), water (13.40 vs. 9.22%), methanol (0.1137 vs. 0.028%), FFA (0.07 vs. 0.29%), ash (2.93 vs. 3.19%), and GE (3,601 vs. 3,625 kcal/kg), current vs. Lammers et al. (2008c),

respectively], which would affect the caloric value of each product.

Even though there were slight differences in crude glycerin composition, averaged among all glycerin products, DE as a percentage of GE was 96.6% (data not shown). This is greater than observed previously (92%; Lammers et al., 2008c), but is similar to the >97% obtained for pure glycerin digested before the cecum reported by Bartelt and Schneider (2002). Likewise, there was no difference in ME as a percentage of GE relative to feedstock source (soybean oil- vs. tallow-based glycerin, $P = 0.75$; soybean oil- vs. yellow grease-based glycerin, $P = 0.56$; tallow- vs. yellow grease-based glycerin, $P = 0.49$; Table 4). The average ME as a percentage of GE for all glycerin products was 85.4%, which is comparable with the 88.4% reported for crude glycerin by Lammers et al. (2008c). Similar to soybean oil and lard, glycerin is utilized well as a source of ME (USP vs. soybean oil and lard, $P = 0.41$) and the purity of glycerin has apparently no effect on this utilization (USP vs. soybean oil-based glycerin, $P = 0.86$).

Because differences in the composition of crude glycerin affect GE and, consequently, ME, we chose to ascertain whether the composition of crude glycerin could accurately predict the ME values experimentally determined in Exp. 1. Using stepwise regression to characterize the relationship of glycerin, water, methanol, fatty acids, and ash content in the crude glycerin to ME yielded the following equation: ME, kcal/kg = 3,580 − (40.52 × % of water) + (48.55 × % of fatty acids), (R^2 = 0.42, P < 0.01). With water and glycerin concentrations moving in opposite directions in the composition of crude glycerin, and because water has no caloric value, removing water from the list of variables subsequently yielded the following equation: ME, kcal/kg = (37.09 × % of glycerin) + (97.15 × % of fatty acids), (R^2 = 0.41; P < 0.01). Because the amount of fatty acids appeared to be a major factor determining the ME in crude glycerin, elimination of the 2 products with an elevated content of fatty acids (IW-NA and USB-GA) from the analysis did not improve the ability to estimate ME from the composition of crude glycerin. Although extreme care was taken in animal feeding, fecal and urine collection, and laboratory analysis, overall variation in the data resulted in a small R^2 estimate of ME from the composition of crude glycerin. In contrast, estimation of GE from the composition of crude glycerin was extremely accurate, where GE kcal/kg = −236 + (46.08 × % of glycerin) + (61.78 × % of methanol) + (103.62 × % of fatty acids), (R^2 = 0.99, P < 0.01). Because there were no differences in ME as a percentage of GE (Table 4), application of the average ME as a percentage of GE (85.4%) to the predicted GE could also be used to predict ME. Applying the above prediction equation for GE and the ME to GE conversion of 85.4% to the data of Lammers et al. (2008c) resulted in GE and ME predictions of 3,805 and 3,249 kcal/kg, respectively, which compare favorably with the actual experimental values of 3,625 and 3,207 kcal/kg, respectively. Likewise, one could apply conversion estimates obtained from complete diets (Noblet and Perez, 1993) to estimate dietary DE, ME, or NE.

A biological approach to calculating the caloric value of crude glycerin could also be used to estimate the GE of crude glycerin based on the GE for glycerin (4,325 kcal/kg; Table 4), oil (9,443 kcal/kg; Table 4), and methanol (5,425 kcal/kg; Bossel, 2003). With these values and the composition of crude glycerin listed in Table 1, the predicted GE concentrations would be 4,311, 3,624, 3,652, 3,687, 3,710, 3,640, 3,191, 3,526, 4,174, 5,763, and 5,335 kcal/kg for USP, AGP, REG-R, REG-WL, REG-MN, ADM-MO, WW-OH, WW-TX, IW-AC, IW-NA, and USB-GA, respectively. These values are highly correlated (r = 0.99) with the analyzed GE values shown in Table 4. Not surprisingly, the slope values obtained by stepwise regression are relatively similar to the biological GE estimates for glycerin (4,608 vs. 4,325, respectively), methanol (6,178 vs. 5,425, respectively), and oil (10,362 vs. 9,443, respectively). As indicated above, one could apply the average ME as a

percentage of GE (85.4%) to these data to predict ME. Applying this approach to the data of Lammers et al. (2008c) resulted in a GE and ME of 3,789 and 3,236 kcal/kg, respectively, which compares favorably with the actual values of 3,625 and 3,207 kcal/kg, respectively, or the regression GE and ME estimates of 3,805 and 3,249 kcal/kg, respectively.

The performance of nursery pigs fed these same diets is presented in Table 5 (Exp. 2). Pigs fed the control diet grew faster (P = 0.02) and were more efficient (P = 0.01) than pigs fed any of the other diets. Because of differences in the ME determined for each complete diet (Exp. 1, data not presented), we assumed it was more appropriate to represent BW gain per megacalorie of intake. Likewise, pigs fed the control diet had a greater BW gain:Mcal of ME intake than pigs fed any other diet (P = 0.01), with no differences between pigs fed any other diet (USP vs. soybean oil and lard, P = 0.81; USP vs. soybean oil-based glycerin, P = 0.46; soybean oil- vs. tallow-based glycerin, P = 0.49; soybean oil- vs. yellow grease-based glycerin, P = 0.89; tallow- vs. yellow grease-based glycerin, P = 0.53). We did not expect differences in G:F because past (Mourot et al., 1994; Kijora et al., 1995, 1997; Kijora and Kupsch, 2006) and more recent studies (Groesbeck et al., 2008; Lammers et al., 2008b; Hansen et al., 2009) have reported similar G:F between pigs fed diets with or without crude glycerin. Only 1 data set has reported a small decline in G:F with crude glycerin supplementation (Della Casa et al., 2009). The reduced G:F in pigs fed all but the control diet could be a function of dilution of nutrients in the diet because in these experiments, the test ingredient was top dressed on the basal diet. For pigs fed crude glycerin, the 9% dilution would have reduced the true ileal digestible Lys to approximately 1.05%, although this still should have been adequate according to the NRC (1998) requirements. Although soybean oil and lard were added at a reduced rate, because oils and fats have 2.25 times the energy of starch (and basically glycerin; Table 4), one would expect that the change in nutrient-to-energy ratio would have had a greater effect on BW gain:Mcal of ME in the diets containing more ME (IW-NA, USB-GA, soybean oil, and lard), but this was not the case. Consequently, we do not have a clear explanation for these data.

Methanol has warranted special consideration in the use of crude glycerin because it is not completely removed at the biodiesel production facility. Methanol is a potentially toxic compound and can elicit a variety of acute and chronic symptoms (Roe, 1982; Medinsky and Dorman, 1995; Skrzydlewska, 2003). Although gastrointestinal disturbances are one of the symptoms of chronic exposure to methanol, there was no apparent effect of dietary methanol on pigs fed IW-NA (3.49% methanol) or USB-GA (14.99% methanol) because no more pigs were removed, relative energy utilization indices (i.e., ME as a percentage of GE) did not differ, and G:F was not decreased compared with pigs fed other glycerin products. In the United States, there is

Table 5. Effect of diet containing crude glycerin, soybean oil, or lard on pig performance, Exp. 2[1]

Item	ADG, kg	ADFI, kg	G:F, kg/kg	BW gain:Mcal of ME[2]
Diet[3]				
Control	0.479	0.683	0.712	212
USP	0.383	0.652	0.584	173
ADM-MO	0.420	0.696	0.602	180
AGP-IA	0.364	0.594	0.609	186
REG-MN	0.436	0.718	0.612	183
REG-R	0.354	0.624	0.564	170
REG-WL	0.342	0.612	0.561	168
WW-TX	0.436	0.702	0.621	186
WW-OH	0.423	0.703	0.607	184
IW-AC	0.394	0.660	0.597	178
IW-NA	0.386	0.637	0.607	174
USB-GA	0.429	0.675	0.631	185
Soybean oil	0.361	0.580	0.620	168
Lard	0.371	0.547	0.676	183
Pooled SE[4]	0.035	0.049	0.024	7.1
Contrast,[5] P-value				
Control vs. all others	0.02	0.47	0.01	0.01
Control vs. USP	0.06	0.65	0.01	0.01
Control vs. soybean oil and lard	0.01	0.05	0.03	0.01
USP vs. soybean oil and lard	0.70	0.14	0.04	0.81
USP vs. soybean oil-based glycerin	0.81	0.91	0.69	0.46
Soybean oil- vs. tallow-based glycerin	0.43	0.39	0.65	0.49
Soybean oil- vs. yellow grease-based glycerin	0.97	0.97	0.95	0.89
Tallow- vs. yellow grease-based glycerin	0.56	0.53	0.77	0.53

[1]Data represent 8 replications of individually penned pigs (4 barrow replicates and 4 gilt replicates, initial BW of 8.70 kg) in 2 groups (25- and 18-d experiments, respectively). There were no 3- or 2-way interactions between group, sex, or diet, or for sex for any criterion measured.

[2]Derived using ADG (g basis) divided by the ME intake (Mcal) obtained from the complete diet in Exp. 1.

[3]Control diet or source of glycerin or lipid: USP = US Pharmacopeia grade glycerin; ADM-MO = Archer Daniels Midland (Mexico, MO); AGP-IA = Ag Processing Inc. (Sergeant Bluff, IA); REG-MN = Renewable Energy Group (Albert Lea, MN); REG-R = Renewable Energy Group (Ralston, IA); REG-WL = Renewable Energy Group (Wall Lake, IA); WW-TX = Westway Feed Products (Houston, TX); WW-OH = Westway Feed Products (Cincinnati, OH); IW-AC = Imperial Western Products acidulated glycerin (Coachella, CA); IW-NA = Imperial Western Products nonacidulated glycerin (Coachella, CA); USB-GA = US Biofuels (Rome, GA).

[4]Pooled SE was calculated by averaging the SE calculated by the GLM procedure (SAS Inst. Inc., Cary, NC) for the variable of interest.

[5]Preplanned contrasts: soybean oil-based glycerin (ADM-MO, AGP-IA, REG-MN, REG-R, REG-WL, WW-TX), tallow-based glycerin (WW-OH), and yellow grease-based glycerin (IW-AC, but excluding IW-NA because of its elevated content of FFA).

no Generally Recognized As Safe regulation or American Association of Feed Control Officials definition listing specifications for crude glycerin use in animal feeds such that specifications for pure glycerin defined under USP and Food and Chemical Codex specifications are used for US Food and Drug Administration guidance. Because methanol content is not specifically listed in the USP or Food and Chemical Codex specifications, the US Food and Drug Administration has decided to address free methanol content under CFR573.640, regulation 21, requiring that the quantity of methanol in methyl esters of higher fatty acids should not exceed 0.015% or a quantity shown to be safe for use in animal diets. In addition to the current experiment, it should be noted that Lammers et al. (2008b) did not report any increased incidence in frequency of lesions associated with methanol toxicity in eye, kidney, or liver tissue in growing-finishing pigs fed 5 or 10% crude glycerin containing 0.32% methanol.

Overall, the data presented herein show that the concentrations of glycerin, fatty acids, and methanol affect the GE of crude glycerin, and because crude glycerin is easily digested and metabolized, it can be used as a vi-

able source of energy in growing pigs. In addition, these data suggest that the amounts of ash and methanol have little to no effect on ME utilization. However, the ME and salt concentration of crude glycerin need to be accounted for in feed formulation, and content of methanol needs to be considered for regulatory reasons.

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